

8.7.2015 Interlab study

WEDNESDAY, 7/8

Petra

Transformed miniprepared D2 to TOP10 just to ensure that the plasmid contained the right construct. Followed transformation protocol. Used 1 ul DNA.

1h incubation (37 C, 230 rpm) was started at 10.57.

Plated 20 ul and 50 ul on chloramphenicol plates.

9.7.2015 Interlab measurement

WEDNESDAY, 8/5

Checked the yesterday's plates for the transformants of D2 under UV-light. Both 20µl and 50 µl chloramphenicol plates contained several GFP-expressing colonies so D2 is working.

22.7.2015 Interlab Measurement study

WEDNESDAY, 7/22

Started restriction for the bricks which are needed for constructing D1 (J23101 & 13504) and D3 (J23117 & 13504):

Markins in tubes: AHD1 R, AHD3 R, AHD4 R and AHD5 R with a current date.

Kept all enzymes and buffers on ice if the protocol didn't expect anything else. Added 2,5ul 10x NEB CutSmart Buffer into the tube (material, dimensions, manufacturer. In order to get 250ng DNA for mixtures, the following volumes of purified plasmids were added to tube shown in Table 1 and the total liquid volume of the tube was balanced to be 25 ul with sterile water, after the enzymes were pipetted. Added 0,5ul of each restriction enzyme following Table 1. Mixed by pipetting and spinned the samples to get reagents in the bottom of tubes.

Incubated samples at 37C for 30 min and inactivated the restriction enzymes at 80C for 20 mins.

Samples stored in -20°C.

Table1

| Sample | DNA (ng/μl) | DNA volume added (ul) | Ion-free water added (ul) | Restriction enzyme used |
|---------------|-------------|-----------------------|---------------------------|-------------------------|
| AHD1 (J23101) | 93,5 | 2,67 | 18,83 | EcoRI & SpeI |
| AHD3 (J23117) | 48,8 | 5,12 | 16,38 | EcoRI & SpeI |
| AHD4 (I13504) | 124,6 | 2,01 | 19,49 | XbaI & PstI |
| AHD5 (pSB1C3) | 25,0 | 10,0 | 11,5 | EcoRI & PstI |